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### Tick-borne virus detection in Caribbean ticks using NGS and high-throughput microfluidic real-time PCR (DOMOTICK Project)

Dr. Emmanuel Albina<sup>3,5</sup>

Dr. Mathilde Gondard<sup>1,3</sup>, Dr. Sarah Temmam<sup>2</sup>, Mrs. Elodie Devillers<sup>1</sup>, Mrs. Valérie Pinarello<sup>3,5</sup>, Dr. Philippe Perrot<sup>2</sup>, Mrs. Rosalie Aprelon<sup>3,5</sup>, Dr. Delphine Chrétien<sup>2</sup>, Dr. Marc Eloit<sup>2</sup>, Dr. Pachka Hammami<sup>3,5</sup>, Dr. Muriel Vayssier-Taussat<sup>1</sup>, Dr. Sara Moutailler<sup>1</sup>

<sup>1</sup> BIPAR JRU, Animal Health Laboratory, ANSES, INRA, Ecole Nationale Vétérinaire d'Alfort, Université Paris-Est, Maisons-Alfort, France

<sup>2</sup> Pathogen Discovery Laboratory, Biology of Infection Unit, Inserm U1117, Institut Pasteur, Paris, France

<sup>3</sup> CIRAD, UMR ASTRE, Petit-Bourg, Guadeloupe, France

<sup>4</sup> IdentityPath Platform, Food Safety Laboratory, ANSES, Maisons-Alfort, France

<sup>5</sup> ASTRE, CIRAD, INRA, Univ Montpellier, Montpellier, France

#### Background

Among hematophagous arthropods, ticks transmit the greater variety of pathogens of public health and veterinary importance whose (re)-emergence is recognized worldwide. Whereas the main human and animal tick-borne pathogens are well characterized in the Northern hemisphere, very few are known concerning the diversity of tick species and tick-borne pathogens circulating within the Neotropical zone of the Americas, especially concerning the Caribbean area.

#### Methods

We previously reported on the results of the DOMOTICK project concerning the detection of tick-borne pathogens in individual Caribbean ticks by using a combination of NGS and high-throughput microfluidic real-time PCRs using Taqman probes (BioMark™ dynamic arrays, Fluidigm Corporation). Here, we focus on interesting viruses detected in these ticks. Total RNA extracted from 588 ticks collected in Guadeloupe and Martinique (*Amblyomma variegatum*, *Rhipicephalus microplus*) were sequenced by Illumina HiSeq.

#### Results

Of 27,544 contigs generated, 1% matched with virus sequences available in GenBank of which 74% aligned with known tick-borne viruses. Out of these tick-borne viruses, four were of particular interest, related to viruses only described once in China in 2015. We detected one novel chuvirus, a circular RNA virus of 11,177 bases with 3 ORFs, with 55.6% of identity with the previously described Changping Tick virus. We also detected a new genotype of another chuvirus with 94.3% of identity with the Wuhan Tick virus, a circular RNA virus of 11,395 bases with 3 ORFs. The third interesting virus is a new genotype of the phlebovirus Lihan tick virus with 95 and 97% of identity on the S and L segments, respectively. The last virus detected is a four RNA-segmented new Jingmenvirus with 80% of identity with the original Jingmen tick virus and Mogiana tick virus. By using the BioMark™ dynamic arrays, we were able to determine the prevalence of these four viruses in individual ticks from Guadeloupe and Martinique. Overall, the four viruses were found in both tick species, but the new Changping-like virus was preferentially found in the *Amblyomma* ticks whereas the three others were preferentially found in the *Rhipicephalus* ticks. The prevalence of the viruses in Guadeloupe and Martinique did not show any bias in terms of geographical distribution.

#### Conclusion

The next steps of these studies will consist in assessing the veterinary and public health risks associated with these four new tick-borne viruses and trying to cultivate some of these viruses for better characterization.

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### The characterisation of hepatitis E virus in UK pigs

Dr. Bhudipa Choudhury<sup>1</sup>

Dr. Sylvia Grierson<sup>1</sup>, Ms. Sarah McGowan<sup>1</sup>, Ms. Charlotte Cook<sup>1</sup>, Prof. Falko Steinbach<sup>1,2</sup>

<sup>1</sup> Animal and Plant Health Agency, Weybridge, United Kingdom

<sup>2</sup> University of Surrey, Guildford, United Kingdom

#### Background

Hepatitis E virus (HEV) infection is widespread in the global pig population. Although clinically inapparent in pigs, HEV infection is the cause of hepatitis E in humans and transmission via the food chain has been epidemiologically established. Studies are ongoing to understand how HEV enters the food chain with pigs/pork products being implicated as a source of infection. Following a 2013 study that investigated prevalence of HEV infection in UK slaughter-age pigs samples indicating the highest viral load (i.e. lowest real time PCR Ct value) were selected for further characterisation, the premise being that these strains were more likely to gain entry to the food chain.

#### Methods

This work was a continuation of an analysis of caecal content samples that had been collected as part of the 2013 Zoonoses in UK Pigs Abattoir Study, a cross sectional study of pigs being slaughtered at 14 high-throughput abattoirs (Grierson et al., 2015). At that time these samples (n=629) along with paired plasma samples had been tested for detection of HEV RNA and antibody. Five samples were subsequently selected for analysis by high throughput sequencing (HTS) based on original test data (Ct value). Four samples (006, 022, 493 and 557) were selected based on lowest Ct values and a fifth sample (090) was selected to enable an assessment of the sensitivity of methodology. These five samples had originated from different farms and from 4 different abattoirs in England.

#### Results

HTS was successfully used to obtain the complete coding sequence from five study samples. A natural in-frame insertion was observed within the HEV hypervariable region in two samples. To interrogate whether this mutation may be the cause of high-level viraemia and faecal shedding as observed in the sampled pigs virus isolation and culture was conducted.

#### Conclusion

Based on viral growth kinetics there was no evidence that these insertions affected replication efficiency in vitro, suggesting as yet undetermined host factors may affect the course of infection and consequently the risk of foodborne transmission. Grierson et al., 2015 Emerg Infect Dis. 2015 Aug;21(8):1396–401.